

---

**Full length research article**

---

**SUSCEPTIBILITY OF SOME SPECIES OF MOSQUITOES TO PERMETHRIN PYRETHROID IN ZARIA NIGERIA.**<sup>1</sup>I. S. Ndams<sup>1</sup>, K. M. Laila<sup>1</sup> and Z. Tukur<sup>2</sup><sup>1</sup>Department of Biological Sciences  
Ahmadu Bello University Zaria, Nigeria<sup>2</sup>Department of Biological Sciences  
Bayero University, Kano, Nigeria

\*(Corresponding author)

[indams@yahoo.co.uk](mailto:indams@yahoo.co.uk)**ABSTRACT**

The susceptibility of *Aedes aegypti* Linnaeus 1762, *Culex quinquefasciatus* Say 1823 and *Anopheles gambiae* Giles 1902 was tested on 0.75 % permethrin pyrethroid in the laboratory. All Larval and pupal stages of the mosquitoes were collected from different localities within Zaria, Nigeria and reared to adulthood in the laboratory. The adults that emerged were tested on 0.75 % permethrin strips using WHO Standard Test Kit. Mortalities were recorded at five minutes interval for 1 hr and maintained for 24hrs post-exposure on 7 % sucrose solution after which a final mortality was recorded. The Knockdown times (KDT<sub>50</sub> and KDT<sub>95</sub>) were determined by Probit Analysis. 100 % mortality was obtained for *Ae. aegypti* and *Cx. quinquefasciatus* after 1hr and 24hr exposures, respectively, whereas *An. gambiae* recorded 92.11 % and 90.91 % mortalities after 1hr and 24hr exposure, respectively. The KDT<sub>50</sub> and KDT<sub>95</sub> did not differ significantly ( $P>0.05$ ) between *Ae. aegypti* and *Cx. quinquefasciatus* but differed significantly ( $P<0.05$ ) with *An. gambiae*. The study shows that *Ae. aegypti* and *C. quinquefasciatus* are susceptible to permethrin whereas *An. gambiae* is resistant to the insecticide in the area.

**Key Words:** Permethrin, *Aedes aegypti*, *Culex quinquefasciatus*, *Anopheles gambiae*, resistance, control, pyrethroid, Nigeria.

**INTRODUCTION**

Resistance to pyrethroid insecticides by mosquitoes is becoming a major public health issue in West Africa (Elissa *et al.* 1993; Chandre *et al.* 1999) where insecticide impregnated bednets are being promoted as a promising means of reducing malaria transmission (Bermejo & Veeken, 1992; Carnevale *et al.* 1992). For treating bednets, pyrethroid compounds are the only insecticides recommended by the World Health Organisation (WHO). They are in general, very effective (with a strong excito-repellant effect on mosquitoes) yet having lower mammalian toxicity than organochlorine carbamate and organophosphate compounds and are less likely to bio-accumulate (Mittal *et al.* 1991).

Permethrin is a chemical widely used as an insecticide. It belongs to the family of chemicals called pyrethroids and functions as a neurotoxin affecting neuron membrane by slowing excess of important sodium ions. In agriculture, permethrin is mainly used on cotton, wheat and maize and is also used to kill parasites on chicken and other poultry. Its uses are controversial since as a broad spectrum chemical, it kills indiscriminately; as well as intended pests; it can harm

beneficial insects including honey bees and small mammals such as mice. It is also used in health care to eradicate ectoparasites such as head lice and scabies and in industrial and domestic settings to control pests such as ants and termites (Wikipedia 2006).

Mosquitoes are vectors of diseases. There are a few reported cases of the resistance of *Culex*, *Aedes* and *Anopheles* to chemical based compounds. However, there is little or no information that points to which of these genera of mosquitoes is resistant to permethrin pyrethroid. The paper reports on the susceptibility of the most abundant mosquitoes in the study area to permethrin pyrethroid. The results are discussed with respect to control strategy of mosquitoes of public health in the area and Nigeria in general.

**MATERIALS AND METHODS**

Larval and pupal stages of four mosquitoes species were collected from breeding sites within Samaru and environs in Zaria. *Aedes aegypti* Linnaeus 1762 was collected from the Ahmadu Bello University (ABU) fountain, *Culex quinquefasciatus* Say 1823 was collected from pools of stagnant water in Samaru village and Areas A residential area of ABU, whereas, *Anopheles gambiae* Giles 1902 was collected from a breeding site near Layin Zomo. All the immature stages were reared to adulthood in the laboratory, during which they were fed with fine powder of fish flakes. The susceptibility test was done as soon as the adults emerged using WHO (1998) Standard Test Kit and procedure. A maximum of 100 female mosquitoes in four replicates for each species were tested.

Accordingly, 0.75% permethrin impregnated paper strips was introduced into 4 exposure tubes and rolled to line with the wall of the tube and fastened into position by a wire clip. The control was lined with plain sheet of paper. A pre-test was performed by carefully introducing 25 female mosquitoes of each species into the four holding tubes with an aspirator and allowed to stand for ten minutes. After which the mosquitoes were transferred into the exposure tubes through a hole on the lid that separated the holding tube and the exposure. The exposure tubes were then set upright with the screen-end up and allowed to stand for 1 hour. Records of mortalities were taken at 5 minutes interval. The mosquitoes were carefully transferred back to the holding tubes and kept for 24 hours during which they were fed with 7% sucrose solution. Records of final mortality were taken after 24 hours. All the dead mosquitoes were removed from the holding tubes and identified using morphological keys of (Gillet 1972; Gillies & Coetzee 1987; Service 1980).

**DATA ANALYSIS**

Mortalities were expressed as percentages (Table 1) while KDT<sub>50</sub> and KDT<sub>95</sub> (Table 2) were determined by probit analysis.

**STATISTICAL ANALYSIS**

Analysis of Variance (ANOVA) was used to compare the mortalities between the mosquitoes species and Least Significant Difference (LSD) was used to separate the means.

## RESULTS

Four mosquito species were collected during the investigation. Of the 238 collected, *Ae. aegypti* and *Cx. quinquefasciatus* had 100 (41.49%) each, while 38 (15.77%) were *An. gambiae*. The 3 *An. pretoriensis* representing 1.24% of the catch encountered were excluded from the analysis.

The results obtained from the susceptibility tests with 0.75% permethrin are presented in Tables 1 and 2.

The result in table 1 shows the mean mortality of the mosquitoes (*Ae. aegypti*, *Cx. quinquefasciatus* and *An. gambiae*). It shows that the knock down time of 35 minutes with 100% mortality was achieved for *Ae. aegypti* and no mosquito survived after 24 hours post exposure.

Similarly, 100% mortalities was achieved for *Cx. quinquefasciatus* with a knockdown time of 30 minutes and all died after 24 hours exposure to the pyrethroid.

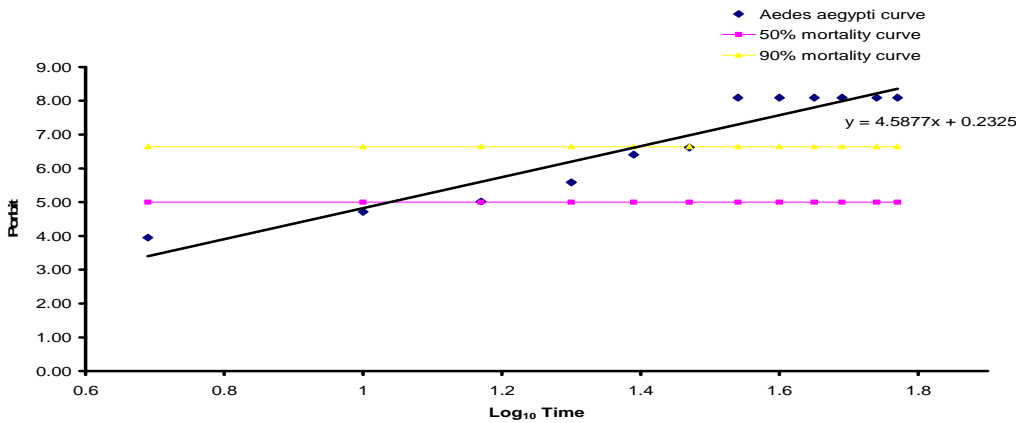


FIG.1: KDT<sub>50</sub> AND KDT<sub>95</sub> of AEDES AEGYPTI.

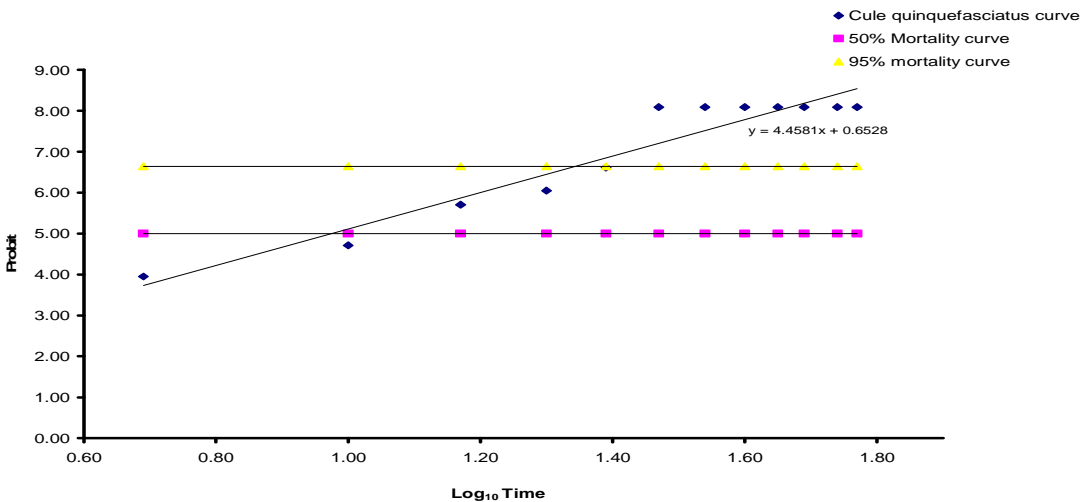


Fig 2: KDT<sub>50</sub> AND KDT<sub>95</sub> FOR C. QUINQUEFASCIATUS.

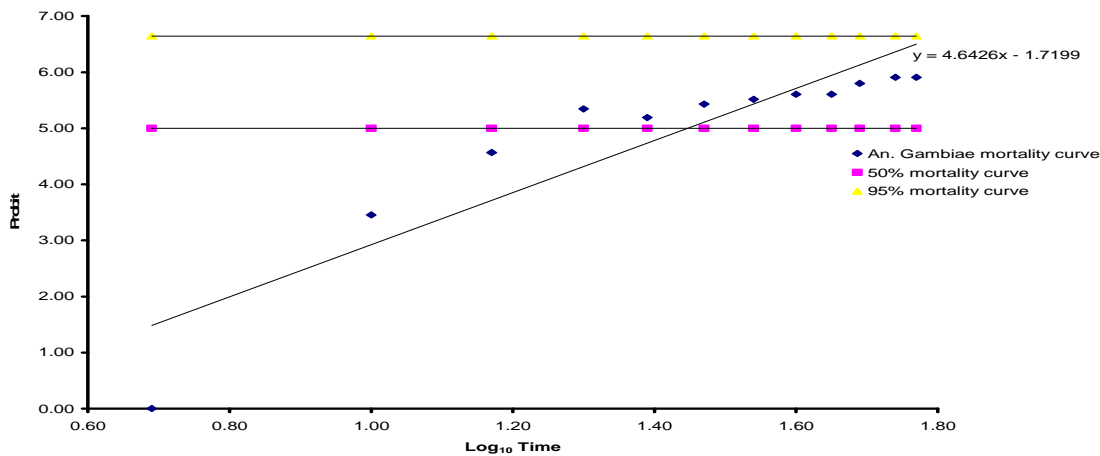


Fig 3: KDT<sub>50</sub> AND KDT<sub>95</sub> FOR AN. GAMBIAE

There was no record of mortality in the control test. *An. gambiae* did not achieve 100% during the 1 hour and 24 hours but 90.91% post exposure observation. Three (3) individuals survived after 24 hr post exposure and were further maintained for 3 weeks after the test to confirmed resistance to the permethrin exposure. Comparing the mortalities caused by the permethrin exposure did not differ significantly between *Ae. aegypti* and *Cx. quinquefasciatus* ( $P > 0.05$ ) but differed significantly between the two species and *An. gambiae* ( $P < 0.05$ ). The mortalities of 3 mosquitoes species differed significantly ( $P < 0.05$ ) with the control test.

Figures 1-3 shows the determination of the KDT<sub>50</sub> and KDT<sub>95</sub> and the result is presented in Table 2 where the mortalities 24 hr post exposure were 100% for *Ae. aegypti* and *Cx. quinquefasciatus* while 90.91% mortality was recorded for *Anopheles gambiae* sl which differed significantly ( $P < 0.05$ ) from the other two species. However, the KDT<sub>50</sub> and KDT<sub>95</sub> as for *Ae. aegypti* and *Cx. quinquefasciatus* are not significantly different ( $P > 0.05$ ) but differed significantly ( $P < 0.05$ ) with *An. gambiae* sl

## DISCUSSION

The investigation revealed that 3 of the 4 species of mosquitoes are prevalent in the study area and was used for the susceptibility tests. They included *Ae. aegypti*, *Cx. quinquefasciatus* and *An. gambiae* s.l. whose prevalences and abundances have been reported (Hanney 1958; Adebote *et al.* 2006). *An. pretoriensis* being reported for the first time in the area was excluded from further analysis because of the small number caught. Further studies on this species are being suggested.

In this study, species are defined as susceptible if they achieve 95% mortality or more and they are defined as resistant if they show less than 95% mortality after exposure to permethrin pyrethroid (WHO 1998; Chandre *et al.* 1999). *Ae. aegypti* and

*Cx. quinquefasciatus* are susceptible to permethrin having achieved 100% mortality respectively, during the 24 hours post exposure, confirming the knock down time effects of 35 minutes for *Ae. aegypti* and 20-30 minutes for *Cx. quinquefasciatus* during which the permethrin pyrethroid caused 100% mortalities, respectively, during the 1 hour bioassay period.

Molta (1979) reported 100% susceptibility of *Cx. pipiens fatigans* and *Ae. aegypti* and suggest that permethrin pyrethroid, though a target insecticide treated bed-nets (ITNs) insecticide against malaria vectors is effective against other vectors of public health importance. *An. gambiae* proved resistance to permethrin pyrethroid having scored 90.91% mortality after 24 hours post exposure and 92.11% mortality observed during the 1 hour test. Furthermore, 3 *An. gambiae* survived after the 24 hour post exposure test which were kept for 2 weeks and fed with 7% sucrose solution. All these results suggest that the *An. gambiae* sl. tested was resistant to permethrin pyrethroid. Awolola *et al.* (2002) reported resistance of *An. gambiae* molecular forms collected from Ikara, a distance of 150km east of Zaria where the current study was conducted. The resistance reported for *An. gambiae* sl in this investigation suggest that permethrin which is a pyrethroid of choice for ITNs in Nigeria and other endemic countries is no longer effective against the malaria vector in the study area. This calls for the use of an alternative pyrethroid or other organophosphates or organochlorine in treating ITNs either single or in combination.

Comparatively, the KDT<sub>95</sub> of *Cx. quinquefasciatus* and *Ae. aegypti* did not differ significantly ( $P > 0.05$ ) but it does ( $P < 0.05$ ) with *An. gambiae* sl suggesting that the behaviours of the mosquito species in the study area may account for the lack of development of resistance to the insecticide.

TABLE 1. MORTALITIES OF MOSQUITO SPECIES AFTER 1HR EXPOSURE TO 0.75% PERMETHRIN PYRETHROID

Control	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00 <sup>a</sup>
<i>Ae. aegypti</i>	14.67	38.67	50.67	72.00	92.00	94.67	100.00	100.00	100.00	100.00	100.00	100.00	84.11 <sup>b</sup>
<i>C. quinquefasciatus</i>	14.67	38.67	76.00	85.33	94.67	100.00	100.00	100.00	100.00	100.00	100.00	100.00	80.22 <sup>b</sup>
<i>An. gambiae</i> l	0.00	6.06	33.33	63.64	57.58	66.67	69.70	72.73	72.73	78.79	90.91	90.91	57.07 <sup>c</sup>

F= 28.9886, F<sub>critical</sub> = 2.8165, P=0.016797, significant: LSD = 10.187  
means with same letters are not significantly different

TABLE 2. PERCENTAGE MORTALITY AFTER 24HOUR RECOVERY PERIOD AND KDT<sub>50</sub> AND KDT<sub>95</sub> AFTER 1HOUR EXPOSURE TO PERMETHRIN INSECTICIDE IN THE MOSQUITO SPECIES

Species	Mortality after 24 hours (%)	KDT <sub>50</sub>	KDT <sub>95</sub>
<i>Ae. aegypti</i>	100	11 <sup>a</sup>	25 <sup>a</sup>
<i>C. quinquefasciatus</i>	100	9 <sup>a</sup>	22 <sup>a</sup>
<i>An. gambiae</i> sl	90.91	28 <sup>b</sup>	63 <sup>b</sup>

Figures with same letters in columns are not significantly different (P>0.05)

*Aedes* and *Culex* are mostly outdoor feeders and breeders whereas *An. gambiae* species is mostly found indoors in close association with human habitations. Similarly agricultural and domestic insecticide have not exerted enough pressure for the development of resistance in *Culex* and *Aedes* but resulted in the development of resistance to permethrin in *An. gambiae* sl.

#### ACKNOWLEDGEMENT

The authors thank the laboratory staff of the Department of Biological Sciences, ABU Zaria for technical assistance.

#### REFERENCES

- Adebote, D. A.; Oniye S. J.; Ndams I. S. & Nache, M, K. 2006. The Breeding of mosquitoes in peridomestic containers and implications in yellow fever transmission in villages around Zaria, Northern Nigeria. *Journal of Entomology*, 3(2):180-188
- Awolola, T. S.; Brooke, B. D.; Hunt, R. H.; & Coetzee M. 2002. Resistance of the malaria vector *Anopheles gambiae* s.s to pyrethroid insecticides in South Western Nigeria. *Annals of Tropical Medicine and Parasitology*, 96:849-852.
- Bermejo, A. & Veeken, H. 1992. Bulletin of the World Health Organisation 70, 293-296.
- Brogdon, W.G & McAllister, J. C. 1998. Insecticide resistance and vector control. *Emerging Infections diseases*, 4(4):1-11.
- Carnevale, P.; Bitsindou P.; Djomande, L. & Robert, V. 1992. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, 86, 362-364.
- Chandre, F.; Darriet, F.; Manga, L.; Akogbeto, M.; Faye, O.; Mouchet J. & Guillet, P. 1999. Status of pyrethroid resistance in *Anopheles gambiae* sl. *Bulletin of the World Health Organisation*, 77:230-234.
- Elissa, N.; Mouchet, J.; Riviere, F.; Meunier, J. Y. & Yao, K. 1993. Resistance of *Anopheles gambiae* s.s. to pyrethroids in Cote d'Ivoire. *Annals de la societe Belge de Medecine Tropicale*, 73:291-294
- Gillet, J. D. 1972. *Common African mosquitoes and their medical importance*. John Swahi, London.
- Gillies, M. T. & Coetzee, M. 1987. *A supplement to the Anophelinae of Africa South of the Sahara*. Publications of the South African Institute for Medical Research No. 55 Johannesburg: South African Institute for Medical Research.
- Hanney, P. N. 1958. The mosquitoes of Zaria province, northern Nigeria. *Bulletin of Entomological Research*, 51:145-171.
- Mittal, P. K.; Adak, T.; & Sharma, V. P. 1991. Acute toxicity of certain Organochloride, Organophosphate, synthetic pyrethroids and microbial insecticides to mosquito fish *Gambusia affinis* (Baird & Girard). *Indian Journal of Malariology*, 28:167-170.
- Molta, N.B. 1979. Preliminary investigation of the insecticidal properties of DBMT on the Mosquito *Culex pipiens fatigans*. B.Sc Project, Department of Biological Sciences, ABU Zaria, Nigeria.
- Service, M. W. 1980. *A guide to medical entomology*. The Macmillan Press Ltd. London.
- Wikipedia, 2006. <http://en.wikipedia.org/wiki/permethrin>.
- World Health Organisation (WHO) 1998. Test Procedures for Insecticide Resistance monitoring in Malaria Vectors, Bio-efficacy and Persistence of Insecticides on treated surfaces. Document WHO/CDS/CPC/MAL/98.12. Geneva